

## **STIC Biotechnology Systems Branch**

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Application Serial Number: 10/598,873  
Source: 1Fwd  
Date Processed by STIC: 9/19/06

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Revised 01/10/06



IFWO

## RAW SEQUENCE LISTING

PATENT APPLICATION: US/10/598,873

DATE: 09/19/2006

TIME: 14:20:12

Input Set : A:\20060914 032301.465 seq list.txt  
 Output Set: N:\CRF4\09192006\J598873.raw

3 <110> APPLICANT: Degussa AG  
 5 <120> TITLE OF INVENTION: Cyanide-tolerant nitrile hydratases  
 7 <130> FILE REFERENCE: 040061  
 C--> 9 <140> CURRENT APPLICATION NUMBER: US/10/598,873  
 C--> 9 <141> CURRENT FILING DATE: 2006-09-14  
 9 <160> NUMBER OF SEQ ID NOS: 14  
 11 <170> SOFTWARE: PatentIn version 3.3  
 13 <210> SEQ ID NO: 1  
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 39 gaa aga gcc tgg gca ttg ttt caa gtc ctc aag agc aag gaa ctc atc  
 40 Glu Arg Ala Trp Ala Leu Phe Gln Val Leu Lys Ser Lys Glu Leu Ile  
 41 10 15 20 25  
 43 ccg gag ggc tat gtc gag cag ctc acg caa ttg gag cac ggc tgg  
 44 Pro Glu Gly Tyr Val Glu Gln Leu Thr Gln Leu Met Glu His Gly Trp  
 45 30 35 40  
 47 agc ccc gag aac ggc gcc cgt gtg gtc aag gcg tgg gtc gat ccg  
 48 Ser Pro Glu Asn Gly Ala Arg Val Val Ala Lys Ala Trp Val Asp Pro  
 49 45 50 55  
 51 cag ttc cgg gca ctg ttg ctc aag gac ggc acc ggc gcc tgc gcc cag  
 52 Gln Phe Arg Ala Leu Leu Lys Asp Gly Thr Ala Ala Cys Ala Gln  
 53 60 65 70  
 55 ttc ggc tac acc ggc ccc cag ggc gaa tac atc gtt gcc ctg gag gat  
 56 Phe Gly Tyr Thr Gly Pro Gln Gly Glu Tyr Ile Val Ala Leu Glu Asp  
 57 75 80 85  
 60 acg ccg acg ctg aag aac gtg att gtc tgc agc ctg tgc tcc tgc acc

Does Not Comply  
 Corrected Diskette Needed

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 in sequence 4,  
 sequence 6,  
 sequence 9  
 51  
 99  
 147  
 (use  
 Eng 13h)

RAW SEQUENCE LISTING  
PATENT APPLICATION: US/10/598,873

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TIME: 14:20:12

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| 62  | 90  |     |     |     |     |     |     |     |     |     |            |            |     |     |     |     | 105  |
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| 66  |     |     |     |     |     |     |     |     |     |     |            |            |     |     |     | 120 |      |
| 68  | ttc | cgc | gca | cgc | ctg | gtc | cg  | gg  | gg  | cg  | ac         | gt         | ct  | cg  | gag | ct  | 435  |
| 69  | Phe | Arg | Ala | Arg | Leu | Val | Arg | Glu | Gly | Arg | Thr        | Val        | Leu | Arg | Glu | Leu |      |
| 70  |     |     |     |     |     |     |     |     |     |     |            |            |     |     |     | 135 |      |
| 72  | ggg | ac  | gag | tt  | ccc | cg  | gac | at  | gt  | gt  | aag        | gt         | tgg | gac | acc | ac  | 483  |
| 73  | Gly | Thr | Glu | Leu | Pro | Arg | Asp | Met | Val | Val | Lys        | Val        | Trp | Asp | Thr | Ser |      |
| 74  |     |     |     |     |     |     |     |     |     |     |            |            |     |     |     | 150 |      |
| 76  | gcc | gaa | agc | cgc | tac | ct  | gt  | ct  | cg  | gt  | agg        | ccg        | gaa | ggc | tca | gaa | 531  |
| 77  | Ala | Glu | Ser | Arg | Tyr | Leu | Val | Leu | Pro | Val | Arg        | Pro        | Glu | Gly | Ser | Glu |      |
| 78  |     |     |     |     |     |     |     |     |     |     |            |            |     |     |     | 165 |      |
| 80  | cac | at  | g   | gaa | gag | cag | ct  | caa | gc  | ct  | gt         | acc        | aaa | gac | gt  | ct  | 579  |
| 81  | His | Met | Ser | Glu | Glu | Gln | Leu | Gln | Ala | Leu | Val        | Thr        | Lys | Asp | Val | Leu |      |
| 82  |     |     |     |     |     |     |     |     |     |     |            |            |     |     |     | 185 |      |
| 84  | atc | ggc | gtc | gcc | ct  | ccc | cgc | gt  | ggc | tga | gaacaacacc | tcatcatcgt |     |     |     |     | 629  |
| 85  | Ile | Gly | Val | Ala | Leu | Pro | Arg | Val | Gly |     |            |            |     |     |     |     |      |
| 86  |     |     |     |     |     |     |     |     |     |     |            |            |     |     |     | 190 |      |
| 88  | tca | act | ccc | gg  | ag  | ttt | gat | ggc | ttt | cac | gat        | ct         | ggc | gg  | ttc | caa | 682  |
| 89  |     |     |     |     |     |     |     |     |     |     |            |            |     |     |     | Met |      |
| 90  |     |     |     |     |     |     |     |     |     |     |            |            |     |     |     | Asp |      |
| 92  | ggc | ttt | gga | aaa | gtc | cct | cac | acc | atc | aac | agc        | ct         | agc | tac | aaa | cag | 730  |
| 93  | Gly | Phe | Gly | Lys | Val | Pro | His | Thr | Ile | Asn | Ser        | Leu        | Ser | Tyr | Lys | Gln |      |
| 94  |     |     |     |     |     |     |     |     |     |     |            |            |     |     |     | 220 |      |
| 96  | gt  | tg  | t   | a   | g   | c   | g   | t   | g   | g   | at         | ct         | gg  | tt  | at  | tc  | 778  |
| 97  | Val | Phe | Lys | Gln | Asp | Trp | Glu | His | Ile | Ala | Tyr        | Ser        | Leu | Met | Phe | Ile |      |
| 98  |     |     |     |     |     |     |     |     |     |     |            |            |     |     |     | 235 |      |
| 100 | gg  | tt         | tt         | tt  | tt  | tt  | tt  | 826  |
| 101 | Gly | Ala | Asp | His | Leu | Lys | Lys | Phe | Ser | Val | Asp        | Glu        | Val | Arg | His | Ala |      |
| 102 |     |     |     |     |     |     |     |     |     |     |            |            |     |     |     | 250 |      |
| 104 | gt  | tc  | g   | tt         | tt         | tt  | tt  | tt  | tt  | 874  |
| 105 | Val | Glu | Arg | Leu | Asp | Val | Arg | Gln | His | Val | Gly        | Thr        | Gln | Tyr | Tyr | Glu |      |
| 106 |     |     |     |     |     |     |     |     |     |     |            |            |     |     |     | 265 |      |
| 108 | cg  | tc  | at  | gc  | ac  | gc  | ac  | ct  | gt  | tc  | g          | ac         | cc  | gg  | gt  | at  | 922  |
| 109 | Arg | Tyr | Val | Ile | Ala | Thr | Ala | Thr | Leu | Leu | Val        | Glu        | Thr | Gly | Val | Ile |      |
| 110 |     |     |     |     |     |     |     |     |     |     |            |            |     |     |     | 285 |      |
| 112 | ac  | c   | g   | tt         | tt         | tt  | tt  | tt  | tt  | 970  |
| 113 | Thr | Gln | Ala | Glu | Leu | Asp | Gln | Ala | Leu | Gly | Ser        | His        | Phe | Lys | Leu | Ala |      |
| 114 |     |     |     |     |     |     |     |     |     |     |            |            |     |     |     | 300 |      |
| 116 | aat | ccc | gcc | cat | gcc | gag | ggc | cgc | ccg | g   | tt         | tt         | tt  | tt  | tt  | tt  | 1018 |
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| 118 |     |     |     |     |     |     |     |     |     |     |            |            |     |     |     | 315 |      |
| 122 | t   | tc  | g   | tt         | tt         | tt  | tt  | tt  | tt  | 1066 |
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| 124 |     |     |     |     |     |     |     |     |     |     |            |            |     |     |     | 330 |      |
| 126 | cac | atc | cg  | at  | cc  | gc  | tc  | gt  | cg  | cg  | g          | aa         | gg  | gg  | gt  | tc  | 1114 |
| 127 | His | Ile | Arg | Met | Pro | Ala | Tyr | Val | Arg | Gly | Lys        | Glu        | Gly | Val | Val | Leu |      |

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| 131 | His Arg Thr Ser Glu Lys Trp Pro Phe Pro Asp Ala Ile Gly His Gly     |     |     |      |
| 132 | 350   | 355 | 360 | 365  |
| 134 | gat gta agc gca gcc cat caa ccc acc tac cac gtc gag ttc gcc gtg     |     |     | 1210 |
| 135 | Asp Val Ser Ala Ala His Gln Pro Thr Tyr His Val Glu Phe Ala Val     |     |     |      |
| 136 | 370   | 375 | 380 |      |
| 138 | aag gac ctg tgg gga gat gcc gcc gat gag ggt ttt gtg gtg gtc gac     |     |     | 1258 |
| 139 | Lys Asp Leu Trp Gly Asp Ala Ala Asp Glu Gly Phe Val Val Val Asp     |     |     |      |
| 140 | 385   | 390 | 395 |      |
| 142 | ctg ttc gaa agc tac ctg gac aag gcc ggc gcg cgcg gtg aac            |     |     | 1306 |
| 143 | Leu Phe Glu Ser Tyr Leu Asp Lys Ala Ala Gly Ala Arg Ala Val Asn     |     |     |      |
| 144 | 400   | 405 | 410 |      |
| 146 | cca tga cagacggcgc ccaggcaagc cgactgcgg tgacggctt ttcgggcttc        |     |     | 1362 |
| 147 | Pro   |     |     |      |
| 150 | ctcggcgccg gcaagaccac cctgctcaac cacatcctgc gcaatcgca aggccctgcgc   |     |     | 1422 |
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| 154 | gatgtcgcgc tgcaccgtgg tcgcgatgag ctgatcgaga tgagcaacgg gtgcacatctgc |     |     | 1542 |
| 156 | tgcaccctgc gcgcgcattt gtcgagcag atcagcatgc tcgcacgcca acagcgtttc    |     |     | 1602 |
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| 204 | gccttttgc gtttctacaa actttttgt ttattttct aaatacattc aaatatgtat      |     |     | 3042 |
| 206 | ccgctcatga gacaataacc ctgataaaat cttcaataat attgaaaaag gaagagtatg   |     |     | 3102 |
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| 210 | tttgctcacc cagaacgcgt ggtgaaagta aaagatgcgt aagatcgtt ggtgcacga     |     |     | 3222 |
| 212 | gtgggttaca tcgaactgga tctcaacagc ggtaagatcc ttgagagtt tcgccccgaa    |     |     | 3282 |
| 214 | gaacgttttc caatgtatgag cactttaaa gttctgtat gtggcgccgtt attatccgt    |     |     | 3342 |
| 216 | gttgacgccc ggcaagagca actcggtcgc cgcatcactt attctcagaa tgacttgg     |     |     | 3402 |

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Output Set: N:\CRF4\09192006\J598873.raw

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| 220 | agtgctgcc   | taaccatgag | tgataaacact | gcggccaact  | tacttctgac  | aacgatcgga  | 3522 |
| 222 | ggaccgaagg  | agctaaccgc | ttttttgcac  | aacatggggg  | atcatgtac   | tcgccttgat  | 3582 |
| 224 | cgttggaaac  | cggagctgaa | tgaagccata  | ccaaacgcac  | agcgtgacac  | cacgatgcct  | 3642 |
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| 228 | cggcaacaat  | taatagactg | gatggaggcg  | gataaaagttg | caggaccact  | tctgcgctcg  | 3762 |
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| 234 | acggggagtc  | aggcaactat | ggtgaacga   | aatagacaga  | tcgctgagat  | aggtgcctca  | 3942 |
| 236 | ctgattaagg  | attggtaact | gtcagaccaa  | gtttactcat  | atatactta   | gattgattta  | 4002 |
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| 240 | aaaatccctt  | aacgtgagtt | ttcggttccac | tgagcgtcag  | accccgtaga  | aaagatcaaa  | 4122 |
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| 248 | caccacttca  | agaactctgt | agcaccgcct  | acatacctcg  | ctctgtaat   | cctgttacca  | 4362 |
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| 254 | cgaacgaccc  | acaccgaact | gagataccca  | cagcgtgagc  | tatgagaaag  | cgcacacgctt | 4542 |
| 256 | cccgaaaggga | gaaaggccga | caggtatccg  | gtaagccgca  | gggtcgaaac  | aggagagcgc  | 4602 |
| 258 | acgagggagc  | ttccaggggg | aaacgcctgg  | tatcttata   | gtcctgtcgg  | gttcgcccac  | 4662 |
| 260 | ctctgacttg  | agcgtcgatt | tttgcgtatgc | tcgtcagggg  | ggcggagcc   | atgaaaaaac  | 4722 |
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| 268 | cgcctgatgc  | ggtatccct  | ccttacgcat  | ctgtcggtt   | tttcacaccc  | cataatgg    | 4962 |
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| 272 | gtacgtgac   | tgggtcatgg | ctgcggcccg  | acacccgcca  | acacccgctg  | acgcggccctg | 5082 |
| 274 | acgggcttgc  | ctgctcccg  | cattcgctt   | cagacaagct  | gtgaccgtct  | ccgggagctg  | 5142 |
| 276 | catgtgtca   | aggtttcac  | cgtcatcacc  | gaaacgcgc   | aggcagctgc  | gtaaaagctc  | 5202 |
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| 280 | gagttctcc   | agaagcgta  | atgtctggct  | tctgataaag  | cgggccatgt  | taagggcggt  | 5322 |
| 282 | ttttcctgt   | ttggctca   | gtgcctccg   | tgtaaggggg  | aatttctgtt  | catggggta   | 5382 |
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| 288 | aaaatcactc  | agggtcaatg | ccagcgtctc  | gtaatacag   | atgttaggtt  | tccacagggt  | 5562 |
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| 292 | gtttccagac  | tttacaaac  | acggaaaccg  | aagaccattc  | atgttgtgc   | tcaggtcgca  | 5682 |
| 294 | gacgtttgc   | agcagcgtc  | gttgcacgtt  | cgctcgctt   | tcgggtattc  | attctgtctaa | 5742 |
| 296 | ccagtaaggc  | aacccgcca  | gcctagccgg  | gtcctcaacg  | acaggagcac  | gatcatgcgc  | 5802 |
| 298 | acccgtggcc  | aggacccaa  | gctgcccgg   | atgcggcc    | tgccgtgt    | ggagatggcg  | 5862 |
| 300 | gacgcgtatgg | atatgttctg | ccaagggtt   | gtttgcgtat  | tcacagtct   | ccgcaagaat  | 5922 |
| 302 | tgattggctc  | caattcttgg | agtgggtat   | ccgttagcga  | ggtgcggcc   | gcttccattc  | 5982 |
| 304 | agtcgaggt   | ggccggctc  | catgcaccgc  | gacgcaacgc  | ggggaggcc   | acaaggatata | 6042 |
| 306 | ggccggccgc  | cctacaatcc | atgccaaccc  | gttccatgt   | ctgcggcc    | ccgcataaaat | 6102 |
| 308 | cgcgtgcac   | atcagcggc  | cagtgtatgc  | agttaggctg  | gtaagagcc   | cgagcgatcc  | 6162 |
| 310 | ttgaagctgt  | ccctgtatgg | ctgtatctac  | ctgcctggac  | agcatggct   | gcaacgcggg  | 6222 |
| 312 | catcccgatg  | ccgcccggaa | cgagaagaat  | cataatgggg  | aaggccatcc  | agcctcgctg  | 6282 |
| 314 | cgcgaacgccc | agcaagacgt | agcccaacgc  | gtcgccgccc  | atgcccggca  | taatggcctg  | 6342 |

## RAW SEQUENCE LISTING

PATENT APPLICATION: US/10/598,873

DATE: 09/19/2006

TIME: 14:20:12

Input Set : A:\20060914 032301.465 seq list.txt  
 Output Set: N:\CRF4\09192006\J598873.raw

316 cttctcgccg aaacgttgg tggcgggacc agtacgaaag gctttagcga gggcgtgcaa 6402  
 318 gattccgaat accgcaagcg acaggccat catcgctcg 6462  
 320 gcccggaaatg acccagagcg ctgcccggcac ctgtcctacg agttgcatga taaagaagac 6522  
 322 agtcataagt gcggcgcacg tagtcatgcc cccgcggcac cggaggagc tgactgggtt 6582  
 324 gaaggctctc aagggcacatcg gtcgacgc 6642  
 326 gcccagtagt aggttggagc cgttgagcac cggccggca aggaatggtg catgcacatcg 6702  
 328 tcaccacaat tcagcaaatt gtgaacatca tcacgttcat ctttccctgg ttgccaatgg 6762  
 330 cccatttcc tgtcagtaac gagaaggctcg cgaattcagg cgcttttag actggctgta 6822  
 332 atgaac 6828  
 335 <210> SEQ ID NO: 2  
 336 <211> LENGTH: 194  
 337 <212> TYPE: PRT  
 338 <213> ORGANISM: Pseudomonas marginalis  
 340 <400> SEQUENCE: 2  
 342 Met Ser Thr Ala Thr Ser Thr Pro Gly Glu Arg Ala Trp Ala Leu Phe  
 343 1 5 10 15  
 346 Gln Val Leu Lys Ser Lys Glu Leu Ile Pro Glu Gly Tyr Val Glu Gln  
 347 20 25 30  
 350 Leu Thr Gln Leu Met Glu His Gly Trp Ser Pro Glu Asn Gly Ala Arg  
 351 35 40 45  
 354 Val Val Ala Lys Ala Trp Val Asp Pro Gln Phe Arg Ala Leu Leu Leu  
 355 50 55 60  
 358 Lys Asp Gly Thr Ala Ala Cys Ala Gln Phe Gly Tyr Thr Gly Pro Gln  
 359 65 70 75 80  
 362 Gly Glu Tyr Ile Val Ala Leu Glu Asp Thr Pro Thr Leu Lys Asn Val  
 363 85 90 95  
 366 Ile Val Cys Ser Leu Cys Ser Cys Thr Asn Trp Pro Val Leu Gly Leu  
 367 100 105 110  
 370 Pro Pro Glu Trp Tyr Lys Gly Phe Glu Phe Arg Ala Arg Leu Val Arg  
 371 115 120 125  
 374 Glu Gly Arg Thr Val Leu Arg Glu Leu Gly Thr Glu Leu Pro Arg Asp  
 375 130 135 140  
 378 Met Val Val Lys Val Trp Asp Thr Ser Ala Glu Ser Arg Tyr Leu Val  
 379 145 150 155 160  
 382 Leu Pro Val Arg Pro Glu Gly Ser Glu His Met Ser Glu Glu Gln Leu  
 383 165 170 175  
 386 Gln Ala Leu Val Thr Lys Asp Val Leu Ile Gly Val Ala Leu Pro Arg  
 387 180 185 190  
 390 Val Gly  
 394 <210> SEQ ID NO: 3  
 395 <211> LENGTH: 220  
 396 <212> TYPE: PRT  
 397 <213> ORGANISM: Pseudomonas marginalis  
 399 <400> SEQUENCE: 3  
 401 Met Asp Gly Phe His Asp Leu Gly Gly Phe Gln Gly Phe Gly Lys Val  
 402 1 5 10 15  
 405 Pro His Thr Ile Asn Ser Leu Ser Tyr Lys Gln Val Phe Lys Gln Asp  
 406 20 25 30  
 409 Trp Glu His Leu Ala Tyr Ser Leu Met Phe Ile Gly Ala Asp His Leu

**VERIFICATION SUMMARY**

PATENT APPLICATION: US/10/598,873

DATE: 09/19/2006

TIME: 14:20:13

Input Set : A:\20060914 032301.465 seq list.txt  
Output Set: N:\CRF4\09192006\J598873.raw

L:9 M:270 C: Current Application Number differs, Replaced Current Application No

L:9 M:271 C: Current Filing Date differs, Replaced Current Filing Date